

### Antimicrobial Activity Of *Waltheria Indica*

\*Zailani, A. Hauwa, #Jada, S, Mahmud and +Wurochekke, U Abdullahi.

Department of Biochemistry, Federal University of Technology, P.M.B,2076 Yola, Adamawa State, Nigeria.  
+[wchekke@yahoo.co.uk](mailto:wchekke@yahoo.co.uk); #[amjnnelushi@yahoo.com](mailto:amjnnelushi@yahoo.com); \*[howwrkoulou@yahoo.com](mailto:howwrkoulou@yahoo.com)

**Abstract:** *Waltheria indica* is used in Nigeria traditional medicine for the treatment of diarrhoea, infertility, skin diseases, gonorrhoea and for relieving pains. Phytochemical analysis revealed the presence of steroids, tannins, saponins, and cardiac glycosides in all parts of the plant; flavonoids were detected only in the leaves and stem, while terpenoids and alkaloids were detected in the leaves only. No part of the plant showed the presence of anthraquinones. Antimicrobial activity of different parts of the plant on *E.coli*, *Pseudomonas aeruginosa* and *Salmonella typhi* showed the leaves having highest activity against *E.coli* and *pseudomonas aeruginosa* with the stem having the lowest activity against the three organisms. Column chromatography of crude extracts of the leaves gave fractions I, II and III that were eluted with ethylacetate/methanol benzene/methanol and acetic acid/methanol respectively. Of these extracts, fraction III showed highest activity against *E.coli* and *Salmonella typhi*. These findings support the traditional use of the plant as an anti diarrhoeal agent.

[Zailani, A. Hauwa, Jada, S, Mahmud and Wurochekke, U Abdullahi. **Antimicrobial Activity of *Waltheria Indica***. Journal of American Science 2010; 6 (12):1591-1594]. (ISSN: 1545-1003). <http://www.americanscience.org>.

**Key words:** *Waltheria indica*, phytochemical analysis, antimicrobial activity, *E.coli*, *Pseudomonas aeruginosa*, and *Salmonella typhi*.

#### 1. Introduction:

The plant *Waltheria indica* belongs to the family *Sterculiaceae*. It is widespread in West Africa (Akobunda and Agyakwa, 1998). Locally, the plant is called 'hankufah' or 'hankubah' in Hausa, 'kafafi' in Fulfulde, 'korikodi' in Yoruba and 'efu-abe in Nupe (Hutchinson and Dalziel 1958; Irvine, 1961). The uses of the plant are diverse; it is used in Northern Nigeria by the Hausas for the treatment of skin diseases, impotence, and infertility, as an aphrodisiac, and as children's medicine at birth and during teething (Mohammed *et al.*, 2007). In the Fulani community, the aqueous extract of the root is used in relieving aches and pains during the 'Sharo' festival. Among the Yoruba, the aqueous extract of the root and stem is used in treating syphilis, internal haemorrhage, and as a restorative during the labours of harvesting (Mohammed *et al.*, 2007). Yerra *et al.*, (2005) investigated the inhibitory effects of the flavonoids isolated from *Waltheria indica* on the production of nitric oxide (NO), tumor necrosis factor alpha (TNF)- $\alpha$  and interleukin 12 (IL)-12, the study supports the use of the plant for the treatment of inflammatory diseases in traditional medicine. The extract of *Waltheria indica* was among the six plants from Northern Cote D'voire that showed promising *in vitro* antibacterial activity against *pneumococcus* including strains resistant to penicillin (Kone *et al.*, 2007). The analgesic activity and/or anti-inflammatory effects have been reported with flavonoids as well as tannins content of the plant (Ahmadiani *et al.*, 1998). Here we present the

antimicrobial activity of the crude and partially separated extract of the plant.

#### 2. MATERIALS AND METHODS

##### *Plant material*

The whole plant of *Waltheria indica* was collected from uncultivated farmlands located at Yola South Local Government, Adamawa State, Nigeria. The plant was identified and authenticated at the department of Biological sciences, Federal University of Technology, Yola, Nigeria.

##### *Micro-organisms*

The micro-organisms used were obtained from the Department of Microbiology, Federal University of Technology, Yola, Nigeria. The organisms include: *E.coli*, *Pseudomonas aeruginosa*, and *Salmonella typhi*.

##### *Extract preparation*

The different parts of the plant namely: leaves, stem and root were removed from the whole plant and air dried separately at room temperature for five days. The dried parts were ground to powder and 50g of each was macerated in distilled water and left overnight. The mixtures obtained were filtered and evaporated using a rotary evaporator.

##### *Phytochemical analysis*

Chemical tests were carried out on the aqueous extract and on the powdered samples using standard procedures to identify the constituents as described by Sofowora (1993); Trease and Evans (1989).

**Antimicrobial activity testing**

The crude extract and individual components were tested using the method of Emeruwa (1982). Wells were made on the surface of 19ml nutrient agar plates previously seeded with 0.1ml of  $10^6$  test organisms. 0.5ml of each extract was aseptically introduced into the wells made. 0.5ml of distilled water was used as control in a separate well. The plates were allowed to stand on the work bench for 30 minutes and were then incubated for 24 hours at 37 °C in an incubator. The presence of zone of inhibition was regarded as the presence of antimicrobial activity. From the inhibition zones, the antimicrobial activity was expressed in terms of average diameter of the zone of inhibition that was measured.

**Separation of crude extract**

The method of Nok *et al.*, (1993) was used. Slurry was prepared by dissolving 30g silica gel in 100ml methanol: water (1:1) and packed in a column (1.5x30cm). The column was loaded with 15ml of the crude extract and sequentially eluted with ethyl acetate/methanol (19:1) benzene/methanol (9:1) and acetic acid/methanol (1:1). The fractions were collected separately and concentrated under pressure using a rotary evaporator.

**Result**

Phytochemical analysis:

The screening of leaves, stems, and roots of *Waltheria indica* using aqueous extract and powdered samples was carried out to determine the presence of tannins, saponins, flavonoids, steroids, alkaloids, terpenoids, cardiac glycosides and anthraquinones. The results are presented in table 1.

**Antimicrobial activity of crude extracts**

The antimicrobial activity of crude extracts of leaves, stem and root were tested on *E.coli*, *Pseudomonas aerogenosa*, and *Salmonella typhi*. The diameter of zone of inhibition is summarized in table 2.

Table2: Diameter of zone of inhibition of crude extracts (mm).

Chemical constituents	leaves	stem	root
<i>E.coli</i>	14	10	11
<i>Pseudomonas aerogenosa</i>	10	10	12
<i>Salmonella typhi</i>	12	8	10

**Antimicrobial activity of the separated eluents of the leaves.**

The recovered extracts from the column weighted 0.5, 0.6 and 0.5g for fractions I, II and III respectively. The antimicrobial activity of the

separated eluents of the leaves is summarized in table 3 below:

Table 3: Antimicrobial activity of the separated eluents of the leaves.

	Diameter of zone of inhibition (mm)		
	Fraction I	Fraction II	Fraction III
<i>E.coli</i>	9	8	10
<i>Pseudomonas aerogenosa</i>	8	12	9
<i>Salmonella typhi</i>	8	7	20

Control (distilled Water) = 4mm

Fraction I=and ethyl acetate/methanol

Fraction II=benzene/methanol

Fraction III= acetic acid/methanol

Table 1: Result of phytochemical analysis of leaves, stem and root of *Waltheria indica*

Chemical constituents	leaves	stem	root
Tannins	++	++	+
Saponins	++	+	+
Flavonoids	++	+	-
Steroids	+	+	+
Alkaloids	+	-	-
Terpenoids	+	-	-
Cardiac glycosides	++	+	+

Key: ++= Highly present +=Present -=absent

**Discussion**

Phytochemical analysis of the extract of the plant revealed the presence of steroids, tannins, saponins, and cardiac glycosides in all parts of the plant. The presence of flavonoids in leaves and stem in this study is in contrast with the opinion of Mohammed *et al.*, (2007) who noted that flavonoids are only present in the leaves with no part of the plant showing the presence of anthraquinones (Table 1). The observed difference could be due to environmental changes where the plants were collected or seasonal changes that could have altered the plant components. It could also have been as a result of changes during extraction and/or storage. Antimicrobial activities of different parts of the plant were tested on *E.coli*, *Pseudomonas aerogenosa*, and *Salmonella typhi*.

The leaves showed highest activity against two organisms- *E.coli* and *Pseudomonas aerogenosa* (Table 1). This could be due to its high composition of cardiac glycosides. Genzolel and Mathel (1982) demonstrated that high composition of cardiac glycosides have been found to inhibit microbial growth and is capable of protection against microbial infection. The phytochemical screening results also

showed the presence of alkaloids and saponins, these classes of compounds have earlier been reported to have antimicrobial activity (Hostettman and Nakanishi, 1979). Therefore, these compounds may be responsible for the observed antibacterial activity of the leaves of *Waltheria indica*.

The root had the highest activity against *Pseudomonas aerogenosa* while the stem had the lowest activity against the three organisms (Table 2). The different components of the leaves i.e. fractions I, II, and III were tested using the same three organisms; fraction III had the highest activity on the organisms, more specifically on *Salmonella typhi*. The result suggests that water is not the most effective solvent for extracting the pharmacologically active compounds and it is a good indication that the most active ingredient of the *Waltheria indica* crude extract is contained in fraction III. It could also mean that the active ingredient maybe different for the different organisms. The fractions reduced the activity of the leaves against the organisms except fraction II, which increased the activity against *Pseudomonas aerogenosa* as compared to the initial crude extract of the leaves (Tables 2 and 3). The observed reduction in activity could have occurred during fractionation.

Gunners (1991) reported that different solvent extracts of some plants may exhibit different pharmacological properties against the same species of microorganisms. Crude extracts may be very active because of synergistic relationship between the components of the plant although sometimes the relationship maybe antagonistic. The observed pattern of antimicrobial activity of crude and various fractions of the plant parts underscores the need to study all parts of the plant before generalization is made on the plant's pharmacological and therapeutic potentials. This is more so because *in vivo*, biotransformation reactions would occur and may likely give rise to new results.

These findings confirm the basis of traditional use of *Waltheria indica* for treating diseases such diarrhoea. The mechanism of action of the constituents of *Waltheria indica* may be difficult to speculate; however, many antibacterial agents may exhibit their action through inhibition of nucleic acid, protein and membrane phospholipids biosynthesis (Franklin *et al.*, 1987). It is probable that the antibacterial agent(s) in the extract of *Waltheria indica* act via some of the above mechanisms. Further studies on the *in vivo* activity, isolation and structural elucidation of the component(s) and toxicological studies of the plant extract are recommended.

**Corresponding Author:**

Zailani, Ahmed Hauwa  
Department of Biochemistry,  
Federal University of Technology, P.M.B 2076, Yola,  
<http://www.americanscience.org>

Adamawa State, Nigeria.

Email: [howwrkoulou@yahoo.com](mailto:howwrkoulou@yahoo.com)

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[editor@americanscience.org](mailto:editor@americanscience.org)

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22/3/2010